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VISUOMOTOR COORDINATION IN THE NEWT (TRITURUS VIRIDESCENS) AFTER REGENERATION OF THE OPTIC NERVE [1]

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THREE FIGURES

It has been shown that normal vision with accurate spatial localization of small moving objects may be recovered in adult urodele amphibians after regeneration of the optic nerve (Stone and Zaur, '40). Whether the normality of the restored vision is due to selective synaptic termination of the regenerating optic fibers on the same central neurons to which they were originally connected, or instead, to a purely functional reorganization after indiscriminate regeneration of the optic fibers has remained a problem.

An explanation on the former basis has been disfavored by abundant evidence that regenerating nerve fibers do not show selective termination (Langley and Hashimoto, '17; Dogliotti, '35; Weiss, '41 a; Kilvington, '41). On the other hand the fixed persistence of maladaptive reactions following 180 degree rotation of the retinal field (Sperry, '43) indicates an extreme rigidity in the functional organization of the visuomotor system of urodeles and would seem to discredit an explanation in terms of relearning or other functionally adaptive processes. Neither explanation has been definitely excluded, however, particularly the latter, since

[1] This research was done at the Harvard Biological Laboratories while the author was on appointment as a Zoology Fellow of the National Research Council.

conditions for adaptive functional reintegration may be much more favorable after optic nerve regeneration than after rotation of the retinal field, with no destruction of the original synaptic relations.

The general problem was approached in the present experiments in the following manner. The eyeball of adult urodeles, Triturus viridescens, was rotated on the optic axis 180 degrees, and the optic nerve was cut and allowed to regenerate. Recovery of normal vision under these conditions would presumably necessitate an adaptive functional readjustment of the central coordinating mechanisms. If, however, regeneration should result not in normal or even confused vision, but in vision with a systematic inversion and reversal that could not be corrected by experience, then it must be concluded that regeneration produces an orderly reestablishment of the original reflex relations by growth processes alone and that functional adjustment plays no part in the recovery. The inclusion by Stone, Ussher and Beers ('37) of two animals with rotated eyes in a group of cases showing normal non-inverted vision after optic nerve regeneration cannot be considered decisive on this point because sufficient data were not given to show conclusively that the vision of these two animals was truly normal.

OPERATIONS AND METHOD

Four differently operated groups of experimental animals were prepared. In group I (twelve cases) the left optic nerve was severed through an incision in the roof of the mouth. The eyeball was allowed to remain in normal position. The dural sheath of the optic nerve was not completely cut but was slit longitudinally. The nerve was pulled out through this slit, broken, and the two stumps tucked back into the sheath. Instead of being cut cleanly, the nerve was teased and pulled apart with fine jeweler's forceps thus precluding any neat coaptation of the ends of individual fibers. This was done intentionally to favor random redistribution of the regenerating axons. The major bulk of the ocular muscles

was excised to prevent movement of the eyeball in case such movement might hinder regeneration. Group I was prepared for the purpose of confirming the previous reports of normal vision after optic nerve regeneration and for use as a control series for the following groups.

In group II (thirty-two cases) the left eyeball was rotated anterodorsally 180 degrees on its optic axis as described previously (Sperry, '43) and allowed to heal in the rotated position. Two days later the left optic nerve was severed and the ocular muscles were excised as in group I. (If the ocular muscles are not excised, their action over a prolonged period tends gradually to pull the eyeball out of the rotated position back toward the normal orientation.) This was the main experimental group. As already indicated, the object of the operation was to find out if regeneration of the optic nerve would lead to normal vision, or to systematically inverted and reversed vision conforming to the new orientation of the retinal field.

Group III (fifteen cases) was operated in the same manner as group II except that, in addition, the right eye was excised a few days after the left optic nerve had been severed. These animals were thus completely blind during the time in which the nerve was regenerating. It was thought that possibly total blindness during this period might weaken the original visuomotor coordinations and so facilitate the establishment of new coordination patterns to suit the revised relations between retina and visual centers.

In group IV (fourteen cases) the left eye was rotated anterodorsally 180 degrees and the left optic nerve was sectioned. This group differed from group II in that both the nerve and its sheath were completely severed with a single transverse cut. Severance of the sheath eliminated any guiding influence which an intact nerve sheath might exert on fiber outgrowth across the gap between the nerve stumps. The distal stump with its sheath was twisted anterodorsally to correspond with the orientation of the eyeball. The proximal stump was twisted in the opposite direction back to its

normal orientation. The two cut ends of the nerve were thus left facing each other 180 degrees out of phase. In addition both stumps were considerably stretched and manipulated with the object of destroying any residual torsional tension on the intervening medium that might tend to guide the regenerating fibers toward their previous pathways in the proximal stump.

The animals of all groups were adult but varied in length from about 5 cm. to about 10 cm. No significant age difference was observed in the results. After operation the animals were kept in a moist chamber for a few days and then returned to large tanks of well water at room temperature. They were fed twice a week on fresh beef and liver.

Tests of vision were made on the tenth day after operation and twice a week thereafter. The unilaterality of visual reflexes in these animals (Sperry, '43) makes it possible to test the function of one eye without blinding the other. The response criteria used in judging vision included compensatory movements of the head to rotation of the visual field, visual localization of small moving objects, and various other visuomotor reactions already described at some length elsewhere (Sperry, '43). These test reactions are clear-cut and quite adequate for determining whether visual perception is normal in character or systematically reversed and inverted.

RESULTS

In the great majority of cases, visual responses to photic stimulation of the left eye reappeared about 30 days after section of the left optic nerve. In nineteen cases, however, return of function was exceptionally delayed and did not appear until about 75 to 95 days after nerve section. These nineteen animals were also exceptional in the following respects. The left eyeball diminished in size until at 3 weeks after operation it was only about four-fifths to two-thirds as large as before operation. The cornea became clouded and the pigmentation pattern of the iris was disarranged. By the time function was restored, the cornea had gradually

recovered its transparency, the pigmentation of the iris had improved considerably, and the eyeball had increased somewhat in size although it was still markedly smaller than in the other experimental animals. Histological study of the retina and optic nerve after return of function showed that the inner nuclear and ganglion layers of the retina were slightly thinner than normal and that there was no localized point in the course of the nerve where the fibers were extremely intertangled as at the point of severance in the other cases. Taken together, the facts indicate (Stone and Zaur, '40) that in these nineteen exceptional animals the retina underwent degeneration and then regenerated from the ciliary margin as described by Stone and Zaur in Triturus after excision and reimplantation of the eyeball. Cutting off the blood supply to the retina in the course of the operations in these cases was probably responsible in large part for retinal degeneration. True retrograde degeneration of the retinal ganglion cells due to section of their axons occurs to a very limited extent, if at all, in these adult urodeles.

One of these nineteen animals was from group I, five of them were from group II, four from group III, and the remaining nine were from group IV. When the visuomotor reactions finally reappeared in these exceptional cases, the responses were essentially like those of the other members of the same groups. Hence further separate treatment of these cases is unnecessary. On the basis of these nineteen animals, however, the experimental results may be extended to include the situation in which degeneration and regeneration of the retina and optic nerve are involved as well as that in which the nerve regenerates by first intention from the point of severance.

When it was ascertained that visual function of the left eye had returned, the right eye was removed in those groups in which this had not been done previously so that the final tests of vision were made in all cases on animals retaining only the left, operated eye.

Visual perception as recovered after optic nerve regeneration was not confusedly blurred as might be expected if the fibers had reconnected at random in the centers without functional readjustment. In all cases an orderly type of vision was recovered so that the animals were able to make specific and consistent localizing responses to small moving objects and compensatory movements of the head to rotation of the visual field.

Recovery with unrotated eye (group I). The reactions to visual stimuli of the animals whose eyeball had been left in normal position were found, on complete recovery, to be indistinguishable from those of normal newts whose right eye had been excised. When an upright drum with vertical stripes was revolved around the newts in a clockwise direction, they moved their heads slowly to the right following the movement of the vertical stripes of the visual field. This photokinetic response did not occur when the drum was rotated in the opposite direction. When a lure consisting of a small piece of meat impaled on the end of a wire was moved outside the glass wall of their aquarium, the newts swam directly toward it from various positions in the aquarium, so long as the lure was visible to the left eye. When the lure was moved inside the aquarium, the newts followed it around for considerable distances, turning in the correct direction when the direction of movement of the lure was changed and snapping at the lure when they came close to it. If the lure was moved quickly over to the right side where it could not be seen, and some distance away so that it could not be detected by vibration of the water, the animals were unable to follow it until by chance they turned again, so that it could be seen with the left eye. In the above tests and in locomotion and other general behavior as well, the reactions of these animals were quite like those of normal control animals from which one eye had been removed.

The fact that small objects were seen discretely and localized in space and that perception of the direction of movement was recovered means that stimulation of different retinal

areas produced specific functional effects and that this specificity of effect was orderly and systematically correlated with the spatial relations of the retinal field. Whether the structural basis for this normal visuomotor coordination is reestablished in the centers by purely growth processes or whether functional adaptation is also an influential factor cannot be determined from these cases in which the retinal field was normally oriented. Rotation of the retina so that restoration of the normal retino-central relations would lead to maladaptive function instead of normal function, however, permitted separation of the two factors and the results obtained in the remaining cases showed that the reestablishment of reflex relations in the visual centers is quite independent of functional adaptation.

Recovery with rotated eye (groups II, III, and IV). All of these cases (except three animals of group II which escaped from their aquarium and were lost) recovered vision which was systematically reversed and inverted conforming to the rotated position of the retina. They displayed definite photokinetic reactions to rotation of the visual field, but the responses were made in reverse direction. When the drum with striped inner walls was revolved counterclockwise about the animals, they slowly turned their heads to the right in the direction opposite to that in which the visual field was moving. When the visual field revolved about them clockwise, no compensatory head movement was made. There was little to distinguish the photokinetic reactions of these newts from those of normal controls except that the movements were made in the reverse direction. When a small moving lure was held above the animals, they turned their heads downward; when it was held below them, they turned upward; when it was held in front of them they turned around and looked behind them; and when it was held behind them and slightly to one side, they started forward. These reactions were definitely not avoidance reactions, but attempts to approach the lure. Spatial localization of small objects was thus consistently erroneous in direct correlation with the retinal rotation.

When swimming or walking on the bottom after recovery of vision, the newts with rotated retinas continually moved in circles. These circus movements were always made in the clockwise direction. When at rest the animals frequently swung the head slowly to the right, then straightened it quickly and again moved it slowly toward the right side. The slow phase of this swaying movement of the head was always toward the right. The experimental cases never swam steadily against the glass walls of the aquarium trying to get through as normal animals and the control animals of group I did frequently. The behavior of the experimental cases after recovery of vision was quite similar in all respects to that which has already been described for newts in which the right eye had been excised and the left eye rotated 180 degrees leaving the optic nerve intact (Sperry, '43). In a few of the cases this was not precisely true because the eyeball during the period of regeneration had gradually pulled out of its operated alignment and had rotated back toward the normal position until in two cases it was nearer 90 than 180 degrees of rotation when function was recovered. In these two cases visual localization was erroneous by 90 rather than 180 degrees. In all cases error of visual localization was directly correlated with the degree of rotation of the retinal field.

Fifteen cases (seven from group II, and four each from groups III and IV) were kept as long as 50 days after recovery of vision. During this period there was no correction of the maladaptive visuomotor reactions. The absence of any functional adaptation in recovery of vision was perhaps most strikingly demonstrated in the cases of group III. These newts were completely blind for at least 27 days and three of them were blind for more than 2 months. When vision was gradually restored after optic nerve regeneration, it came back in all cases reversed and inverted and remained so thereafter unmodified by experience.

During the regeneration period while these animals were blind they managed to locate food and to get about the aquarium fairly well by relying upon the remaining senses.

When vision returned, however, they began to swim and walk in circles and to collide with obstacles much more frequently and forcibly than when they had been blind. It took these newts longer to locate food after vision had been recovered than when blind. In general the animals appeared to be at a greater disadvantage with reversed vision than with no vision at all. The absence of any functional reorganization under the conditions of these experiments extends the previous evidence of the lack of functional plasticity in the basic coordination patterns of the visuomotor system in Triturus (Sperry, '43).

Chromatophoric reactions. Rotation of the retinal field 180 degrees without severing the optic nerve has been shown to result in a color change in these animals such that their dorsal surface becomes distinctly lighter in color (Sperry, '43). If the afferent mechanisms controlling this melanophoric response had been correctly restored by nerve regeneration in the present experiments, the animals with rotated eyes should have become lighter in color when regeneration was completed and the normal eye removed. Distinct color changes of this kind were not noticed. Although some of the newts became slightly lighter in color, none of them became as light as did the newts whose eyes had been rotated with the nerve intact. Many of the animals remained as dark as they had been before operation. Apparently restoration of the relations for chromatophore control by way of the hypothalamus was inconsistent and only partial at best. The explanation must await further investigation.

Anatomical analysis. Twelve cases were prepared for microscopic examination. These included six cases from group II in two of which retinal degeneration had occurred and two each from groups I, III, and IV. Bodian's ('36) silver proteinate method was used with the minor modifications for Triturus suggested by Singer ('42). The optic nerve was straightened and slightly stretched before fixation by pulling the eyeball far enough out of the orbit so that the eyelids

could be pushed under it. The nerves were sectioned longitudinally in different planes at 10 μ.

In the ten cases in which the nerves had regenerated by first intention from the point where they were cut, the point of severance was clearly marked by an extreme intertangling of the fibers which took a very crooked and devious course through this region. Figure 1 shows a section through the scar region of a case from group IV. It is interesting that the discrete functional effects of different excitations sent through the optic nerve after retinal stimulation were not destroyed by passage of the excitation patterns through this very tangled snarl of nerve fibers in the scar region. Proximal to this point the fibers were comparatively straight as in the normal nerve (fig. 2). After crossing the gap to the proximal stump, the regenerating axons are apparently guided along the parallel framework left by the degenerating nerves. The appearance of the regenerated fibers beyond the scar region through the chiasma along the central tracts to the optic tectum was not noticeably different from that of normal animals. Details of the complicated terminations within the tectal alba itself could not be followed, of course, in this type of preparation.

There was no marked difference between cases from different groups except that the neuromatous tangle marking the point of severance was most extensive in the animals from group IV in which the nerve sheaths had been completely cut. No distinct decussation was found in any of the cases to indicate that individual regenerating fibers from the rotated distal nerve stump had entered specifically their original degenerated pathways in the normally oriented proximal stump. Because of the extreme intermixing of fibers in the scar region, however, it is impossible to rule out decisively such selective regeneration on the basis of the histological appearance alone. Indications of occasional but not profuse bifurcation of nerve fibers were present in the scar region.

In those animals in which the eyeballs had shrunken in size and return of vision had been long delayed, due apparently

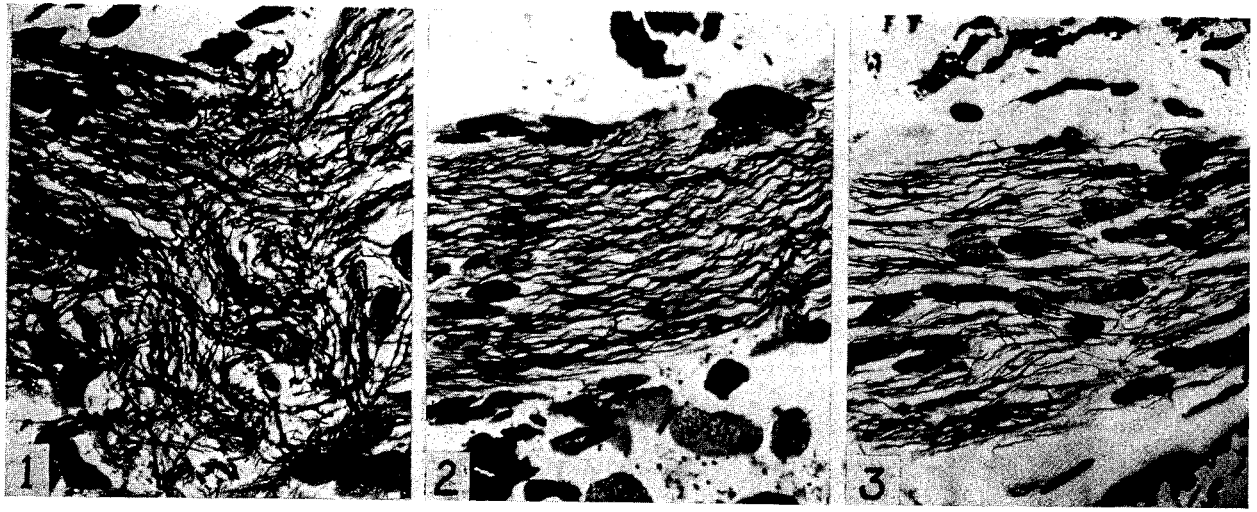

Fig. 1 Photomicrograph of a section through the scar region of a regenerated optic nerve. × 400.

Fig. 2 Photomicrograph of a section through a more proximal region of the same regenerated nerve shown in figure 1. × 500.

Fig. 3 Photomicrograph of a section of regenerated optic nerve of a case in which the retinal elements as well as the proximal stump of the nerve degenerated (see text). The nerve is shown at the point where it passes through the optic foramen. × 500.

to degeneration of the retina, no distinct scar region was present at the point where the nerve had been severed. The nerve tracts were not so compactly arranged as in the other cases and considerable intermixing and crossing of the fibers was apparent throughout the course of the nerve from retina to chiasma. The irregular interweaving of the fibers in one of these cases is shown in figure 3 at the point where the nerve passes through the brain case, a point where the connective tissue sheath remained intact and where the nerve was well protected from any possible mechanical disturbance at the time of operation. The contortion and intermixing of fibers was more extensive in these cases than in the regions proximal and distal to the scar in the other animals, due probably to disintegration of the old nerve framework. In their studies of retinal degeneration and regeneration in adult Triturus, Stone and Zaur ('40) found that the old nerve stumps underwent complete disintegration before the new nerve fibers grew out from the regenerated retina. The irregular pattern of the regenerated fibers in the present cases gave further indication that the course of individual axons was not predetermined by old pathways.

DISCUSSION

Visual perception after regeneration of the optic nerve in these urodeles was not a blurred confusion. Small objects were seen discretely and localized in a consistent manner. Perception of the direction of movement was also systematically restored. Clearly the original functional values or so-called "local signs" of all the various retinal loci were recovered without confusion after the fibers had reconnected with the visual centers of the brain. This occurred even though the retinal field had been rotated 180 degrees so that restoration of the original functional effects was not only without adaptive value but was distinctly disadvantageous to the animals. The resultant reversal of responses to visual stimuli, moreover, remained rigidly fixed thereafter without any correction by functional reorganization. The results

demonstrate that functional adaptation is not influential in determining the character of the recovered visual perception. It must be concluded instead that the original reflex relations are reestablished in the centers in an orderly manner by the regeneration process itself.

This leaves the interesting problem of exactly how the systematic structural associations for organized visual perception are laid down by purely growth processes. Very likely the factors operating here in regeneration are similar in nature to those involved in pre-functional differentiation of precise reflex relations in embryonic development, a process about which extremely little is yet known.

With regard to this problem, one conclusion can be drawn directly from the present experiments, namely, that ganglion cells of the retina including their optic fiber axons must differ intrinsically from each other according to their locations in the retinal field. Fibers arising from different retinal loci must be distinguished in the centers by some differential factor in order to obtain recovery of specific spatial localization of small objects and perception of the direction of movement. In the absence of functional adjustment, no such specialization of function could be regained if the regenerating fibers were all identical.

The results require that ganglion cells of each quadrant of the retina have intrinsic physico-chemical qualities distinguishing them from the ganglion cells of the other retinal quadrants. Ganglion cells located in different regions of any one quadrant must likewise have distinctive physico-chemical properties correlated with their specific functional values. It is easiest to conceive of this retinal cell differentiation as being orderly and continuous so that the difference between cells located far apart across the retina is greater than that between cells which are nearer together. The development of such a condition is readily interpreted embryologically in terms of a polarized, field differentiation of the optic cup. The retinal field would thus become, in respect to cellular differentiation of the ganglion layer, a true "field" in the

physical sense of the term. Presumably if rotation of the retinal field or its anlage were carried out at a sufficiently early stage of development, the polarity of retinal differentiation would be regulated so that normal vision would be recovered. The original polarity of the retina is evidently restored after degeneration and regeneration in these adult urodeles probably through organizing influences spreading from the ciliary margin of the retina and perhaps also from other structures of the eyeball which resist degeneration.

The constitutional specification of the optic tract fibers revealed by these experiments belongs in the same category as the similar biochemical specificities of peripheral nerves already well demonstrated in amphibians by the extensive experiments of Weiss ('36, '42). In accordance with Weiss' Resonance Principle ('28, '36) this nerve fiber specificity may determine the quality of the excitatory impulses which the different kinds of fibers discharge and the selective sensitivity of different neuron types to the various kinds of impulses. The basic functional organization of the nerve centers is thus conceived to depend not upon a specificity of fixed neuron connections, but upon the emission of qualitatively distinct excitatory agents and a selective receptivity to these discharges permitting functional precision despite profuse, unrestricted neuropil interconnections. (See also Schiff, 1896; Hering, '13; Head, '20.) This conception of the role played by the nerve fiber specificities may well be the correct interpretation in the case of the present results. It would account for the orderly recovery of precise functional relations despite random, indiscriminate termination of the optic fibers in the visual centers.

Another possible interpretation of the manner in which this nerve fiber specificity determines functional relations, proposed by the author ('41) must also be considered. Namely, that these neuron specificities act primarily in the growth process in regeneration and in embryonic development to influence the formation of central synaptic connections. The specification of nerve fibers by their end-organs is considered to be an important factor in regulating the establishment of

proper anatomical associations between peripheral and central neurons. Accordingly in the case of the present experiments the optic fiber specificity induced by differentiation of the retina may permit the various kinds of regenerating fibers to form specific central connections owing to differential affinities and incompatibilities between the ingrowing axons and the central neurons. Orderly reestablishment of central synaptic relations in this way would also account for the functional phenomena and this interpretation would not demand any drastic revision of the prevalent connectionist theory of nervous integration.

At present no crucial data are known that would favor or eliminate either of these possible interpretations of the effect of nerve fiber specification and the situation remains problematic (Weiss, '41 b). The idea of qualitatively distinct excitatory impulses and selective receptivity, quite applicable in the case of limb coordination where a discrete set of muscles is involved, or in the case of the corneal reflex with a single receptor specificity (Weiss, '42), becomes somewhat more difficult to apply to the visual system where the functional specificities of the retina are extremely numerous and intergraded. For this reason and because the Resonance Principle and its implications have already been fully outlined elsewhere (Weiss, '28, '36), further consideration is here devoted to the possibility of explaining the present results in terms of the reestablishment of specific anatomical associations rather than in terms of specific nerve energy and resonance phenomena, although it is fully recognized that this latter possibility is by no means excluded.

That the individual axons found their own degenerated counterparts in the proximal stump and simply followed these pathways to their old terminals in the brain does not appear probable for reasons already mentioned. Moreover the same sort of prefunctional organization must occur in embryonic development without any pre-existing degenerated pathways to guide the ingrowing axons. Another possibility is that fibers from the same region of the retina tend to adhere to

each other during regeneration because of preferential surface affinities (selective fasciculation) so that the whole optic tract is normally oriented with an orderly internal arrangement as it approaches the visual centers. The interweaving and crossing of fibers, not only in the scar region, but also throughout the course of the nerve in those cases in which retinal degeneration occurred does not support this assumption. Also, Herrick ('41 a, '41 b) has shown that in other urodeles there is normally extensive intermixing of fibers in the central part of the optic nerve with a segregation at the chiasma which does not correspond to retinal quadrants and is different from that where the nerve leaves the retina. This segregation at the chiasma may be due more to mechanical conditions at the chiasma at the times when the different types of fibers arrive than to selective fasciculation based on physico-chemical properties of the fibers. Ströer's ('40) contrasting claim that the fibers in Triturus taeniatus maintain their original segregation throughout the optic tracts from retina to tectum is based on a descriptive study of serial sections without selective staining or selective operational analysis and must await confirmation by more critical methods. It appears most probable from the present evidence that only general factors, in large part mechanical, guide the regenerating optic tract as a whole to its terminal central stations, and that the distribution of individual fibers as they approach the optic tectum is largely random.

It is questionable, however, that the optic fibers must necessarily approach the optic tectum with any orderly internal arrangement in order that particular fibers may terminate on particular tectal neurons. Herrick ('41 b) describes conditions in the optic tectum of Necturus as follows: "It is clear that the dendritic spread of many of the tectal neurons is so wide as to cover the entire width and thickness of the tectum, thus permitting synaptic contact with all types of afferent fibers regardless of their local arrangements." Also he finds that the optic fibers terminate in the alba by wide arborizations. Although the dendritic spread is

probably not so extreme in the less primitive tectum of Triturus, it is difficult to believe that sufficient specificity of connections could be attained by mere guidance of the approaching fibers toward particular regions of the tectum without further discrimination. In view of the wide arborization of the axon terminals, the dendritic spread, and the small size of the urodele tectum, invading axon tips would be able to make contact with any tectal neurons without having to grow very far regardless of the exact point at which the axons happened to enter the optic area. Moreover since fibers from each retinal area are very numerous — Herrick ('41 a) estimates about 6000 fibers in the optic nerve of Amblystoma — and each axon undergoes arborization in the tectum, the chance of fibers from the various retinal loci reaching proper tectal neurons is quite favorable when considered in statistical terms. Given the possibility of extensive contacts among the tectal neurons, it is only necessary, in order that appropriate synaptic linkages be restored, that the tendency to cease further growth and form synaptic endings be preferential according to specific affinities between any invading fiber and the various central neurons which it encounters.

Thus even though the fibers approach the centers with a random distribution, the possibility of their forming specific associations through discrimination and selection within the terminal stations themselves is not excluded. The alba of the optic tectum where the majority of optic fibers terminate consists of a dense feltwork of tectal dendrites and axons of afferent fibers from other nuclei (Herrick, '25 a, '41 b). On entering this dense tangle of nerve filaments, the invading fibers do not form intimate synaptic endings on any and all neuron processes they happen to encounter. Termination is selective at least to the extent that synapsis is restricted largely if not entirely to dendrites only. The more subtle specificities existing between different types of regenerating fibers and between different types of dendrites may result in further selectivity of termination among the dendrites.

The specificity of central connections so established may depend merely upon a discrete distribution of synaptic junctions. It may also depend upon systematic quantitative differences in the excitatory capacities of the synaptic relations formed. This explanation of the reestablishment of retino-central relations in the adult would be applicable also to the original prefunctional organization of these relations in embryonic development.

The foregoing interpretation assumes, of course, a differentiation of the neurons of the optic tectum, not merely in respect to their functional relations with lower centers, but also in physico-chemical properties that are capable of influencing growth processes previous to and irrespective of the adaptability of the functional results. According to the hypothesis of end-organ induction of central connection specificity, it is presumed that the central neurons become specified originally through a series of successive induction steps starting with the motor neurons whose specification is imposed upon them by differentiation of the musculature. This is consistent with the rather general rule, to which the visuomotor system is no exception (Herrick, '37), that in early embryogenesis the neuromuscular units develop first, after which they become linked in appropriate patterns by subsequent differentiation of connecting pathways so that maturation of the motor and central adjustor system is already sufficiently advanced to permit organized responses to stimuli as soon as, and even before, the sensory connections are made. Twitty ('32) has shown that invasion of the optic fibers into the tectum permits, or directly stimulates, further proliferation of neurons in the optic zone. This appears, however, to be a rather generalized effect resulting merely in additional neuroblasts. It is only after the indifferent neuroblasts have migrated from the germinal layer and begun to send out axon and dendritic filaments that the specific influences affecting functional reflex relationships with which we are here concerned become effective. Specificity resulting in formation of proper synaptic associations is

probably imposed upon these additional neuroblasts at an advanced stage of maturation by neurons already differentiated.

The regulative influence of peripheral differentiation on the formation of central reflex relations, as conceived here in explanation of the present results on the visuomotor system, may have wider application to the general problem of prefunctional organization of the central nervous system. According to this theory the synaptic relations between motor, adjustor, and sensory neurons are laid down systematically in development in conformance with and as a result of the end-organ connections acquired by the peripheral nerves (Sperry, '41). Embryonic differentiation of the end-organs by inducing corresponding physico-chemical specification of the peripheral nerve cells thereby transmits the effects of peripheral differentiation into the primary sensoria and motoria of the central system. Thereafter, by consecutive induction of intermediate and higher associations, the primitive inherent reflex relations of the nervous system attain a hierarchical organization based directly upon the primary connections with the periphery.

The inherent development of precise reflex associations in the centers and their arrangement to suit peripheral connections without aid of functional adjustments becomes more comprehensible in these terms. The scheme allows for considerable developmental variation in the absolute topography of nerve connections both intra-central and peripheral without affecting functional relations and so accounts on a connectionist basis for normal function of the anomalous variations that appear not infrequently in the nervous system of any species.

Rather than autonomous central self-differentiation of coordination patterns (Weiss, '41 b) this hypothesis would emphasize peripheral induction of central reflex relations. The great importance of peripheral organ influence on the early proliferative development of the nerve centers has been amply demonstrated (Detwiler, '36; Szepsenwol, '36).

That the peripheral influences might be more detailed and specific in the later stages of differentiation, such that end-organ relations affect the formation and organization of central connections has often been supposed (Herrick, '25 b; Larsell, '31; Nicholas and Barron, '35; Szepsenwol, '36; and others). Functional or implicit discharge of the developing neurons and the resultant ionic fields set up about the fibers has generally been presumed to be the organizing factor. The assumption that it is instead a common induction effect of cell growth and differentiation such as is instrumental in regulating developmental organization of other tissues would simplify rather than complicate the problem, and is more readily subject to experimental test. The possibility that central synapsis may be governed from the periphery in this manner appears to warrant further investigation in view of the evidence furnished by the present experiments.

SUMMARY

1. In seventy adult newts the left optic nerve was severed in ways designed to favor random and abnormal reassortment of regenerating fibers. This was accompanied in fifty-eight of the cases by rotation of the left eyeball on its optic axis through 180 degrees. Fifteen of the latter animals were made completely blind by excision of the right eye in addition to severance of the left optic nerve.

2. The optic nerve regenerated by first intention in fifty-one of the animals and vision was restored in about 30 days. In the remaining nineteen cases the retina underwent degeneration and recovery of vision was exceptionally delayed to about 85 days.

3. The character of the recovered vision conformed in all cases to the orientation of the retina. It was normal in the twelve animals in which the eyeball had been left in normal position. It was systematically inverted and reversed in the fifty-eight cases in which the retinal field had been rotated 180 degrees. Although the reversed vision with the consequent maladaptation of visual reactions proved to be a

greater handicap to the animals than total blindness, it remained uncorrected by functional reorganization.

4. Clearly the original functional values of various retinal points were systematically restored after regeneration. Since this occurred despite a maladaptive effect for the animals, it is concluded that the central reflex relations are reestablished in an orderly manner by the growth process itself without aid of functional adaptation.

5. Regenerating optic fibers arising from different retinal loci must be distinguished from each other in the centers according to the relative location of their ganglion cell bodies in the retinal field, probably by differential physicochemical properties imposed on them by a polarized differentiation of the retina in development. The experiments thus extend the demonstration of peripheral nerve specificity in amphibians (Weiss, '42) to include the fibers of the optic tract.

6. The manner in which the optic fiber specificity influences the reestablishment of functional relations in the visual centers can be accounted for either on the basis of the Resonance Principle or on the hypothesis that formation of central synaptic associations is regulated by developmental inductive effects from the periphery. The latter alternative is discussed and the possibility of its more general application to some of the problems of prefunctional organization of central reflex relations is indicated.

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